Antioxidants in aqueous extract of *Myristica fragrans* (Houtt.) suppress mitosis and cyclophosphamide-induced chromosomal aberrations in *Allium cepa* L. cells. *Journal of Zhejiang University-SCIENCE B (Biomedicine & Biotechnology)*, 12(11):915-922.

Akinboro, A., Kamaruzaman B. M., Mohd Zaini, A., Ahmad, S.O., Tang, H.Y. Siti Marina, M. (2012). Mutagenic and antimutagenic assessment of methanol leaf extract of

Myristica fragrans (Houtt.) using *in vitro* and *in vivo* genetic assays. *Drug and Chemical Toxicology*, Vol. 35 (4), 412-422.

Babu, T.D., Kuttar, G., & Padikkla, J. (1995). Cytotoxic and antitumour properties of certain

taxa of umbelliferae with special reference to *Centella asiatica* (L.) Urban. *Journal of Ethnopharmacology* 48, 53-57.

Schimmer, O., Kruger, A., & Paulini, H. (1994). An evaluation of the 55 commercial plant

extracts in the Ames mutagenicity test. *Pharmazie*. 49(6): 448-451.

Seema, C.C., & Meena, V. (2012). Cytotoxic and genotoxic effects of *Centella asiatica*

extract in the cultured human peripheral blood lymphocytes. *Research in Pharmacy*, 2(4): 31-40.

Siddique, Y.H., Ara, G., Beg, T., Faisal, M., Ahmad, M., & Afzal, M. (2007). Protective role of *Centella asiatica* L. extract against methyl methanesulphonate and cyclophosphamide induced genotoxic damage in cultured human lymphocytes. *Recent Progress in Medicinal Plants* 19, 369–381.

Siddique, Y.H., Beg, T., & Afzal, M. (2008). Antigenotoxic effect of ascorbic acid against cypropterone acetate induced genotoxicity in cultured mammalian cells. In: *Recent Trends in Toxicology*, YH. Siddique (Eds), Transworld Research Network,Trivandpuram, Kerala, India. 85-94.

Siddique, Y.H., Beg, T., & Afzal, M. (2009). Effect of Centella asiatica L. Extract against ethinylestradiol induced genotoxic damage on cultured human lymphocytes. Biomedical Research, 20 (2): 141-147.

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INDUCED GENETIC VARIABILITY IN SESAME (*SESAMUM INDICUM*(L) USING FAST NEUTRON IRRADIATION AND SODIUM AZIDE.

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ABSTRACT

Induced Genetic variability's in sesame was studied. Variety kenena-4 was exposed to fast neutron irradiation (FNI) from an Am-Be source with a flux of $1.5\times10^{4}ncm^{-2}s^{-1}$, using doses of 0,4,8,12 and 16µsv and also treated with sodium azide using doses of 0.0%,0.2%,0.4%,0.6%,0.8%. Plant height, survival percentage, length of petiole, number of leaves per plant and leave surface area were observed for quantitative character .There were significant differences (at $p<0.05$) in kenana-4 at different doses of fast neutron irradiation and sodium azide with most of the parameters used. Both radiation and chemical showed negative correlation with most of the parameters used.

Key words: Mutagenesis, FNI, sodium azide, sesame.

INTRODUCTION

Sesame is considered to be the oldest oilseed crop known to man. The crop has been domesticated well over 5000 years (Bisht *et al.*,1998). It belongs to the family Pedaliaceae and genus *Sesamum*. The genus consist of about 36 species out of which the commonly recognized is *Sesamum indicum* L. (Falusi, 2006). *Sesamum indicum* is very drought tolerant. It has been called a survivor crop because of its ability to grow where most plants fail. The crop is believed to have originated from Africa where the greatest diversity of the genus sesame and its family Pedaliaceae is present (Falusi and Salako, 2003). Some of the local names of the crop in Nigeria are ("Ridi" Hausa) ("Ishwa" Tiv), ("Gorigo" Igbira), ("Eeku" Yoruba) and ("Doo"Jukun) (Falusi *et al*, 2001)

Currently it is cultivated in the tropical and sub tropical region of Africa, South America, North America and Asia principally for its seeds which contains about 50-52 % oil, 17-19 % Protein and 16-18 % carbon hydrate (Falusi and Salako, 2003).It is an annual plant growing to 50 - 100cm (1.6 to 3.3 ft)tall, with opposite leaves 4 -14cm (1.6 - 55in) long with an entire margin :they are broad lanceolate, to 5cm(2in) broad at the base of the plant, narrowing to just 1 cm (0.4in) broad on the flowering stem . The flowers are yellow, tabular, 3 to 5cm (1.2 to 2.0m) long, with four lobed mouths. The flower may vary in colour with some being white, blue or purple. Sesame fruit is a capsule, normally pubescent, rectangular in section and typically grooved with a short triangular beak. The length of the fruit capsule varies from 2 to 8 cm. Its width varies between 0.5 to 2cm, and the number loculi from 4 to 12. The fruit naturally splits opens (dehisces) to release the seed by splitting along the septa from top to bottom or by means of two apical pores, depending on the varietal cultivar. Sesame seeds are small, about 3to4millimeter long by 2millimeter wide and 1millimeter thick. The seeds are ovate, slightly flattened and somewhat thinner at the eye of the seed(helium)than the opposite end with the weight of the seed between 20to 40 milligrams. Sesame is grown primarily for it's oil-rich seeds. The oil is used locally for cooking as well as for medicinal purposes such as the treatment of ulcers and burns. The stem and the oil extracts are equally used in making local soup.The products are locally processed and utilized in various forms. Principally among the products are "KATUN RIDI" and "KANUN RIDI". After oil has been extracted from the seeds, the cake is made into "Kuli Kuli" which together with the leaves are used to prepare local soup known as "MIYAR TAUSHE". Artificial induction of mutation is of scientific and commercial interest as it is one of the methods used in improving the growth and yield of economic plants. It provides raw materials for the genetic improvement of economic crops (Adamu et al., 2004). Although various mutagens were known to induce mutation in plants, this work has made use of fast neutron and sodium azide in inducing genetic variability through mutagenesis to improve both the quality and quantity of sesame. The aim of the present study is to induce mutation through the use of various concentrations of sodium azide and fast neutron in sesame (S. indicum L. Var. Kenana-4) to improve the quality and quantity of the plants.

MATERIALS AND METHODS

The study was carried out at the experimental garden; Centre for Preliminary and Extra-mural Studies, Federal University of Technology, Minna, Niger State, Nigeria.

The seeds were obtained at National Cereal Research Institute (NCRI) Baddegi, Niger State.

The seeds were irradiated with fast neutron at the Centre for Energy and Research training (CERT), Ahmadu Bello University Zaria, Kaduna state, Nigeria. Kenana 4 was subjected to different doses of fast neutron. The variety was divided into five equal parts and exposed to 0, 4,8,12 and16µsv.

Sodium azide was used to treat the seeds, there were five different concentrations, 0%,0.2%,0.4%,0.6%,0.8%. Sodium azide was diluted to the required concentration by using distilled H2O.Seeds were soaked in the H2O for six hours to initiate Biochemical reaction. The chemical reaction is found to be affected by the frequency and spectrum of mutagens depending on the type of cell division during the process of germination. If the chemical treatment is synchronized with DNA synthesis stage (G1, s and G2) then we get better results.

The presoaked seeds were put in flask and Sodium azide was added and left for eight hours. Usually the quantity of Sodium azide 10 times the volume of the seeds. Intermittent shaken was given to ensure uniform exposure of the chemicals. The chemical was drained after the treatment time is over. The seeds were washed immediately not less than 30mins.

Two factors were involved; the variety and irradiation for physical, variety and sodium azide for the chemical. Factorial design was adopted with two (2) plants per pot with a total of 15 combinations per plot. The arrangement used was randomized block design with thirty (30) pots per block (figure 1).The experiment was replicated in three making a total of 90 pots for physical and 90 pots for chemical. Ten seeds were planted per pot (i.e. five per hole in a pot).Three weeks after planting; each pot was thinned to two plants per pot. A total of eight (8) pots for each treatment combination were used.

The following data were taken during the period of study;

Plant height at 2, and 4weeks after planting and at maturity: The distance from ground level up to the terminal bud on main axis of a plant in cm using metre rule, length of petiole(cm) using metre rule, leaf surface area in cm^2.Survival rate 21 days after planting: this was taken in percentage. The result of this research was subjected to analysis of variance (ANOVA) to show whether there were significant differences among the morphological parameters and yield parameters. Duncan multiple range was used to separate the means. The survival rate, flowering percentage and the spearman rank correlation was used to show the relationship between the treatments and parameters.

RESULTS

For the effect of fast neutron irradiation kenana 4 variety at 2 weeks, 0usv assumed the highest with a mean of 6.87a, Followed by 8usv, 12usv, 16usv and the least was recorded in 4usv with the mean of 5.67a and there were no significant difference observed in the various doses at $p<0.05$, but there was a negatively very weak correlation(-0.148). At 4 week 12usv showed the highest plant height with the mean of 25.73a, then 0usv, 4usv, 8usv, and the least was recorded in 16usv with a mean of 20.93b. There were significant difference observed between 12usv and all other doses at $p<0.05$. There was a negatively weak (-0.287) not significant correlation between the doses and the plant height However, at 6 week 12usv assumed the highest height (70.25a) than other doses, while the least was recorded in 16usv. With the mean of (53.60b). There were significant difference observed between 12usv and the other doses (0usv, 4usv, 8usv, 16usv) at $p<0.05$. But there was a positively weak correlation (0.100) and not significant. For the effect of sodium Azide on plant height at 2 weeks variety kenana 4 treated with 0.2% and 0.8% were significantly different from 0.4%, 0.6% and 0.0% (controls) with the means of (6.45a and 6.20a) respectively while the lowest was 0.6% with the mean of (3.80b).The correlation was a weak positive correlation (0.238) and not significant.At 4th week 0.6% recorded the least plant height with the mean of 12.64c, while 0.0% (control) has the highest mean (23.78a) followed by 0.2%, 0.4% and 0.8%. 0.0% and 0.2% were statistically different with the other doses at $p<0.05$. The correlation was a strong negative correlation (-0.757) and was not significant.At 6th week, 0.2% had the highest mean of (58.29a) followed by 0.0%, 0.8%, 0.6% and 0.4% recorded the lowest mean of (45.36b). A modest negative correlation (-0.629) not significant was recorded. 0.2% and 0.0% were statistically different from the other doses at $p<0.05$.

For the effect of fast neutron irradiation (FNI) on the number of leaves per plant kenana 4, 16usv assumed the highest (11.10a) and the least recorded 4usv(9.80a). But there were no statistical difference in all the doses at $p<0.05$. There was a strong positive correlation and not significant (0.827).

For the effect of sodium Azide on the number of leaves per plant in kenana 4, 0.2% has the highest number of leaves (27.30a) and the least was recorded in 0.0% (control) with the mean of (10.20c) 0.2% and 0.4% were statistically different from the other doses 0.8%, 0.6% and 0.0% at $p\leq0.05$. The correlation was a weak positive correlation (0.278).

Kenana 4, radiated at 12usv had the longest petiole (5.10a) followed by 4usv, 8usv, 0usv and the least was 16usv with the mean of (2.18c). There were statistical difference between 12usv and the other doses at $p<0.05$. The correlation was a negative weak correlation and not significant. For the effect of sodium Azide, kenana 4 treated with 0.0% had longest petiole (2.90a) the least was recorded in 0.6% with the mean of (1.11b). There were statistical difference between 0.0% and the other doses at $p\leq0.05$. The correlation was a negative modest correlation and not significant.

For the effect of fast neutron irradiation (FNI) kenana 4, 0usv had the highest leave surface area(42.18a) and the least was 16usv(25.61c). There were statistical difference in all the doses at $p<0.05$. There was a weak negative correlation (-0.417) and not significant

For the effect of sodium Azide in kenana 4, 0.0% had the highest leave surface area (42.18a) and the least was recorded in 0.6% with the mean of (27.11b) there were statistically different between 0.0% and all the other doses at p≤0.05. The correlation was a weak negative correlation (-0.315) and not significant.

For the effect of fast neutron irradiation on survival percentage, 16usv and 8usv in kenana 4 performed better (53% and 50%) than the control 0usv (45%). For the effect of sodium azide on survival percentage in kenena 4 0.8%,0.2% and 0.6% (93%,73%and 50%)performed better than the control(45%).

Table 1: Some morphological parameters of the Kenana-4 variety at different doses

TREATMENT	PLANT HEIGHT 2 WEEKS	4 WEEKS	6 WEEKS	NO. OF LEAVES PER PLANT	LENGTH OF PETIOLE	LEAVE SURFACE AREA
FAST NEUTRON						
Ke0	6.87±1.76a	23.78±1.74ab	57.30±14.99ab	10.20±1.22a	2.90±0.96bc	42.18±12.63a
Ke4	5.67±1.42a	23.71±3.80ab	58.89±17.7ab	9.80±1.54a	3.84±1.32b	38.18±4.79ab
Ke8	6.70±1.68a	21.22±2.83b	61.50±17.10ab	10.90±1.59a	3.45±0.84b	37.43±23.57ab
Ke12	6.63±1.35a	25.73±3.45a	70.25±20.93a	10.90±1.72a	5.10±1.55a	30.95±2.50ab
Ke16	6.16±1.89a	20.93±2.57b	53.60±13.83b	11.10±1.37a	2.18±0.50c	25.61±9.84c
SODIUM AZIDE						
Ke0	3.90±0.68b	23.78±5.32a	57.30±14.99a	10.20±1.22c	2.90±0.96a	42.18±12.63a
Ke2	6.45±1.37a	22.60±5.39a	58.29±15.47a	27.30±7.74a	2.23±0.92ab	38.70±4.85ab
Ke4	4.29±1.18b	21.97±5.73ab	45.36±10.80b	25.00±7.87a	1.09±0.16b	39.93±22.63a
Ke6	3.80±1.15b	12.64±5.06c	48.60±7.67ab	16.60±10.37bc	1.11±0.32b	27.11±2.93b
Ke8	6.20±1.12a	17.80±3.88b	51.05±13.07ab	21.60±11.22ab	2.39±3.37ab	32.05±8.56ab

*Values are mean±SD. Values followed by the same letter(s) within the same row do not statistically differ at the 5% level according to DMRT

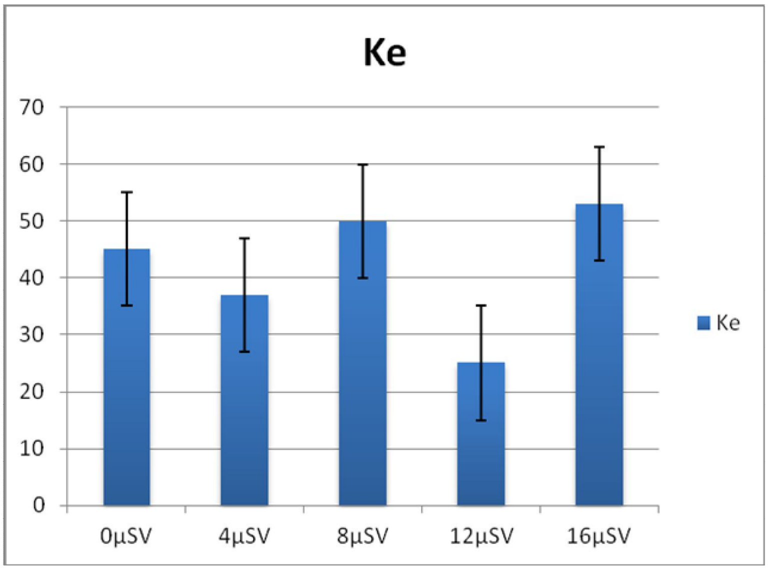

Figure 4.1: Survival Percentages of Kenana-4 at different doses of FNI

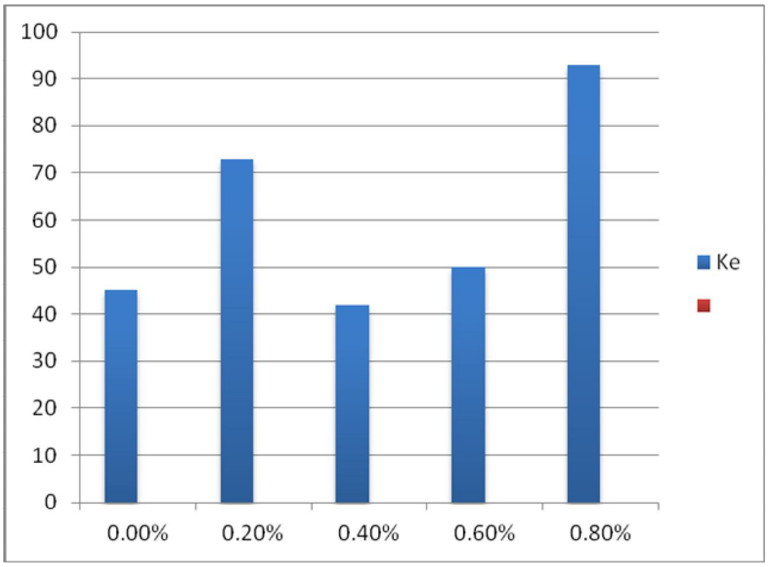

Figure 2: Survival Percentages of kenana-4 at different concentration of sodium azide

DISCUSSION

Mutation induction through the use of different concentrations of sodium azide and fast neutron irradiation has proved vital in inducing variability that could be exploited in the improvements of sesame growth and yields. It is therefore the origin of genetic variability as suggested by Tamarin (1999). The mean increase in plants heights at maturity of the sesame variety induced by sodium was due to the alteration of their genome integrated by environmental signals as reported by Uno et al. (2001); probably by increasing the rates of cellular division and expansion at their meristematic regions. This is also in agreement with the findings of Hoballah (1999) who reported increased in plant heights of sesame due to radiation mutagenesis; but is in contrast to the findings of Anandakumar and Sree- Rangasamy (1995) and Maluszynski et al. (2001) who independently reported decrease in plant height due to induced mutation in rice and other cereals. The increase in leaf number and internodes length with decrease in the concentrations of colchicines was in agreement with the findings of Hoballah (1999) who reported increased in leaf number and internodes length among sesame mutants due to gamma irradiation. The increase in the leaf area of sesame due to colchicines means an increase in the surface area for gaseous ex-change which consequently affects the photosynthetic process. This agrees with the work of Maluszynski et al. (2001) who reported increase in the leaf area among Zea mays mutants due to irradiation. Artificial induction of mutation through the use of sodium azide proves vital in the improvement of genetic variability in sesame. Certain concentrations of sodium azide (0.2 through 2.0mM colchicines concentration) have the potentiality of inducing variability that could be used in the improvement of the yield of sesame.

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REFERENCES

Adamu, A. K., Chung, S. S., and Abubakar, S. (2004): The effect of ionizing radiation (Gammarays) on tomato (s.n.). *Nigerian Journal of Experimental Biology,* 5(2):185-193

Anandakumar, C. R, and Sree-Rangasamy, S. R. (1995): Heterosis and selection indices in rice. *Egyptian Journal of Genetics and Cytology,* 14:123-132.Bisht, I.S., Mahajan, R.K., Loknathan, T.R. &Agrawal, R.C. (1998). Diversity in Indian sesamecollection and stratification of germplasmaccessions in different diversity groups. *Genetic Resource and Crop Evolution* 45(4), 325-335.Falusi, O. A. (2006). Estimation of natural cross pollination in two species of the genus *Sesamum*(pedaliacea), *Production Agriculture and Technology* 2(2):61-65.ISSN:07945213.

Falusi, O.A. and Salako, E.A. (2003). Inheritance studies in wild and cultivated *Sesamum* L. Speciesin Nigeria. *Journal of Sustainable Agriculture*.22 (3):75-90.

Falusi, O.A., Salako, E.A. and Ishaq, M.N. (2001). Interspecific hybridization between *Sesamumindicum* L. and *Cerathothecasesamoides*Endl.*Tropicultura*, P.127.

Hoballah, A. A. (1999): Selection and Agronomic evaluation of induced mutant lines of sesame. In: Induced Mutations for Sesame Improvement IAEA-TECDOC, IAEA,Vienna, pp 71# 84.

Maluszynski, M., Szarejko, I., Barriga, P., and Balcerzyk, A. (2001): Heterosis in crop mutant crosses and production of high yielding lines, using doubled haploid systems.*Euphytica*, 120: 387-398.

Tamarin, R.H. (1999): Principles of Genetics, 6th ed, WCB/McGraw Hill.NY pp1-684.

Uno, G., Storey, R. and Moore, R. (2001): Principles of Botany. Mc Graw Hill New York 1 550pp.